

THE HSFA9 PROGRAM: INTERACTION BETWEEN TWO HEAT-SHOCK FACTORS FOR TRANSCRIPTIONAL ACTIVATION OF SEED sHSP GENES

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We will show that two heat-shock factors, HaHSFA4 and HaHSFA9 synergistically activate sHSP gene transcription in bombarded embryos and leaves from sunflower. HaHSFA4 was recently cloned by yeast two-hybrid interaction using a mutagenised form (which is transcription-inactive) of HaHSFA9 as bait. We observed in planta interaction between HaHSFA4 and HaHSFA9 by bimolecular fluorescence complementation (BiFC); this will be confirmed by GST pull down using plant extracts. The BiFC experiments also showed homomeric interactions for HaHSFA9 and HaHSFA4. In addition, BiFC indicated a differential cellular localization of these two HSFs: the heteromeric interaction between HaHSFA4 and HaHSFA9 could be observed only in the nucleus. As homomers, HaHSFA9 would be mainly localized in the nucleus, whereas HaHSFA4 is partitioned between the nucleus and the cytosol. By confocal microscopy using GFP and DsRed2 fusions, we have confirmed the inferred localization for HaHSFA4, HaHSFA9, as well as nuclear co-localization of both HSFs. We will discuss mechanistic explanations for the observed synergism. The planned combination of HaHSFA4 and HaHSFA9 -in transgenic plants- may improve seed longevity and desiccation tolerance, beyond what already obtained using HaHSFA9 only.